THE COMPARATIVE DISTRIBUTION OF TWO CHROMATOPHOROTROPIC HORMONES (CDH AND CBLH) IN CRUSTACEAN NERVOUS SYSTEMS

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Introduction

It was demonstrated by Brown (1933) that sea-water extracts of the crustacean central nervous organs contained material having a definite and characteristic effect upon certain chromatophores of the body. The nervous organs were the only tissues of the body other than the eyestalks, with their included sinus glands, that yielded such a chromatophorotropically active substance, thus suggesting that the former possibly contained a source or sources of normal, color-changing hormonal material. In the shrimp, *Palaemonetes*, injection of extracts of the nervous system were shown to bring about a rapid blanching of dark-colored specimens through concentration of the red and yellow pigments within the chromatophores, an action similar to that which could be induced by extracts of the sinus gland of the eyestalk.

Similar activity of the nervous system was described by Hosoi (1934) for *Penaeus japonicus* and by Hanström (1937) for *Penaeus brasiliensis*. Knowles (1939) found that extracts of the central nervous system of *Leander adspersus* caused concentration of the white pigment within that species. Concentration of white pigment by extracts of central nervous system was also reported for *Cambarus* by Brown and Meglitsch (1940) who worked with the chromatophores in isolated pieces of integument. Sinus gland extracts had an antagonistic action upon this pigment, thus proving that the sinus glands and nervous system did not yield exclusively identical chromatophorotropic substances.

Evidence that the central nervous organs contained sources of hormones normally involved in the adaptive color-changes of *Palaemonetes* was presented by Brown (1935) who found that any vigorous stimulation of the cut ends of the optic nerves in darkened eyestalkless specimens would induce a blanching characteristic of that following injection of extracts of central nervous organs. Koller (1930) had also observed comparable responses of eyestalkless *Crago* but did not at that time consider the central nervous organs to be a source of the active material.

More convincing evidence for the production of a normal chromatophorotropic hormone in the crustacean nervous system was presented by Brown and Ederstrom (1940). Their observations concerned the reactions of the particularly sensitive melanophores in the telson and uropods of the shrimp, *Crago*. Amputation of the eyestalks of a white-adapted animal brought about, within 3–6 minutes, a complete dispersion of black pigment in the melanophores giving the animal a "black-tailed" appearance. The condition persisted for about an hour whereupon the pigment returned to its former concentrated state, the latter condition typically lasting for several days. Brown and Ederstrom found that the black pigment could be caused

to disperse again by stimulation of the eyestubs or by the injection of extracts of the circumoesophageal connectives. Upon more extensive experimentation they concluded that the mid-region of the connectives, including the connective ganglia, contained the origin of the *Crago* tail-darkening hormone (CDH) involved here. The results of these investigators were confirmed and extended when Brown and Wulff (1941) gave evidence for a second chromatophorotropic principle within the central nervous system, namely a *Crago* body-lightening hormone (CBLH) by describing that strong stimulation of the eyestubs simultaneously darkened the telson and uropods and lightened the remainder of the body, an action duplicated by injection of extracts of the central nervous system as a whole. It was shown that these two actions were due to two separable principles in that injection of ethyl-alcohol extracts of the nervous system gave only body-lightening action, the tail-darkening principle remaining in the alcohol-insoluble residue, and, that mild stimulation of the eyestubs of eyestalkless animals produced both tail-darkening and body-darkening. Brown and Wulff speculated that CDH was, in the absence of CBLH, a general body-darkening principle. This hypothesis was more specifically set forth and given experimental support by Brown (1946) who clearly demonstrated the source of this darkening principle to lie, not in the circumoesophageal connectives proper, but in the minute tritocerebral commissure interconnecting the connectives immediately posterior to the oesophagus. Injection of sea-water extract of this commissure in various experiments produced in every case tail-darkening but various degrees of either body-lightening or body-darkening. The variable effects upon the body seemed reasonably explained in terms of varying concentrations of an antagonistic body-lightening principle.

In the following experiments a survey was made of the effects of sea-water extracts of the central nervous systems of thirteen species of higher crustaceans representing the *Isopoda*, *Natantia*, *Astacura*, *Anomura*, and *Brachyura* upon *Crago* color-change. The distribution of both the *Crago* tail-darkening hormone, CDH, and the *Crago* body-lightening hormone, CBLH, was considered. We have concerned ourselves primarily with the presence or absence of each substance within the central nervous systems and, when the hormones are present in a particular species, with a survey of the relative concentrations of the principles within the parts containing the hormone in question.

Experiments and Results

The experiments to determine the distribution of CDH and CBLH were conducted in the following manner. Animals for use in assaying the concentration of active principles in extracts of nervous tissue were first prepared. The eyestalks of a number of *Crago septemspinosus*, ranging from 3–6 cm. in length, were amputated by means of a sharp scalpel and the eyestubs cauterized with an electric cautery needle. No animals were used for assay purposes until at least twelve hours following this operation, at which time they could best be described as possessing mottled black and white bodies and light telson and uropods (see Fig. 1*A*, control).

A relatively simple but effective method was used in the preparation of central-nervous-system extracts. The donor of the nervous tissue first had eyestalks removed and stubs cauterized in the same manner as described above for *Crago*. The dorsal portion of the exoskeleton was then cut away. After removing surrounding

viscera and muscles the nervous organs were removed under a dissecting microscope by carefully severing the nerves about the brain, thoracic and abdominal cords and gently lifting the entire system out of the animal. Particular caution was observed in the removal of the circumoesophageal connectives so as to prevent any damage to the tritocerebral commissure. The nervous system was then placed in a watchglass containing a small amount of sea-water and divided by means of a sharp scalpel into the desired portions which usually comprised brain, connectives, thoracic cord, and abdominal cord.

FIGURE 1. *A*. Darkening of eyestalkless *Crago* following injection of a sea-water extract of the abdominal nerve cord of *Homarus* (conc. = 1 cord/0.5 ml. sea-water). The two specimens on the left are two uninjected ones used for a control. The injections for the animals on the right were made 15 min. before the photographs were made. *B*. Lightening of eyestalkless *Crago* following injection of a sea-water extract of the circumoesophageal connectives of *Uca* (conc. = 3 pr. conn. to 0.2 ml. sea-water). The two specimens on the right were injected 8 minutes before the photographs were made.

Following this procedure the organs were transferred to individual glass mortars where excess sea-water was removed and the tissues allowed to dry partially. The tissue was then triturated with a measured amount of sea-water varying in quantity with the different species from 0.1–0.5 cc. per portion depending upon the size of the nervous system as a whole. In some cases, such as that of *Idothea,* it was necessary to use the parts of several nervous systems in the preparation of each extract in order to obtain adequate concentration and amount for assay. All extracts were centrifuged for three minutes at approximately 3,500 R.P.M. and the supernatant liquid of each injected into the dorsal musculature of the abdomen of at least two test-animals prepared as described above. The amount of extract injected into each varied with the size of the test-animal, but was normally between

TABLE I

Responses of eyestalkless Crago to injection of extracts of various portions of the central nervous system of other crustaceans. No. of cases signifies the number of donors

			Tail-darkening Time (min.)							Body-lightening ⊖ or darkening ⊕ Time (min.)						
Species	Organ	No. cases	0	5	10	15	30	45	60	0	5	10	15	30	45	60
Homarus	Brain	7	0.0	3.3	3.6	3.9	3.7	3.7	1.6	0.0	0.0	+0.7	+2.0	+2.6	+1.9	+0.5
	Connectives	8	0.0	1.9	2.3	2.3	2.3	2.1	1.4	0.0	−1.4	−1.0	0.0	+0.6	+0.4	0.0
	Thoracic cord	8	0.0	1.6	2.1	2.2	2.8	2.7	2.5	0.0	−0.8	−0.3	+0.4	+2.8	+2.8	
	Abdominal cord	2	0.0	1.0	1.5	2.5	3.0	2.5	1.5	0.0	+3.0	+3.5	+4.0	+4.0	+4.0	+4.0
Cambarus	Brain	10	0.0	3.4	3.4	3.4	2.6	1.3	0.4	0.0	+0.8	+1.2	+1.7	+0.9	+0.4	+0.2
	Connectives	10	0.0	2.5	2.9	3.2	2.2	0.9	0.3	0.0	−1.7	−0.9	−0.6	−0.1	+0.1	+0.3
	Thoracic cord	10	0.0	2.7	3.1	3.4	2.9	2.1	1.3	0.0	−1.9	−1.7	−0.9	+0.1	+0.3	0.0
	Abdominal cord	10	0.0	2.6	2.8	2.8	2.5	1.4	1.0	0.0	+0.8	+1.2	+1.4	+1.1	+0.6	+0.2
Upogebia	Brain	7	0.0	1.3	2.3	2.3	2.2	0.8	0.0	0.0	−2.0	−1.7	−1.5	−0.4	−0.2	0.0
	Connectives	6	0.0	0.2	0.6	0.0	0.0	0.0	0.0	0.0	−2.3	−2.6	−2.0	−1.7	−0.4	−0.2
	Thoracic cord	7	0.0	1.5	2.1	2.7	2.5	2.0	1.4	0.0	−0.2	+0.7	+1.2	+0.8	+0.5	+0.2
	Abdominal cord	7	0.0	1.0	1.4	1.4	1.2	0.9	0.4	0.0	−0.6	−0.6	−0.3	0.0	0.0	0.0
Pagurus	Brain	8	0.0	0.4	0.5	0.4	0.2	0.2	0.0	0.0	−1.6	−1.6	−1.1	−0.5	−0.1	0.0
	Connectives	8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	−1.8	−1.6	−1.0	0.0	+0.1	+0.1
	Thoracic cord	8	0.0	1.9	2.9	3.1	2.4	1.4	0.2	0.0	−1.3	−1.3	−1.3	−0.5	+0.2	0.0
Emerita	Brain	8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	−1.5	−0.6	−0.5	−0.2	0.0	0.0
	Connectives	8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	−1.4	−0.6	−0.2	0.0	0.0	0.0
	Thoracic cord	8	0.0	1.0	1.6	1.6	1.0	0.5	0.1	0.0	−0.9	−0.6	−0.1	−0.1	0.0	0.0
Libinia	Brain	7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	−2.6	−2.8	−2.0	−1.0	−0.3	0.0
	Connectives	7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	−1.7	−1.8	−1.7	−0.4	−0.1	0.0
	Thoracic cord	7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	−1.7	−1.7	−1.8	−0.9	−0.4	0.0

0.025 and 0.04 cc. Sea-water injected or uninjected controls were observed simultaneously with all test-animals.

Observations of the color changes in both body and tail were taken at five-minute intervals up to fifteen minutes and at fifteen-minute intervals thereafter. The degree of darkness of the tail or body was described within the range, + 1 to + 4, the number + 4 representing the maximum extent of darkening and the number + 1, the minimum observable one. In a similar manner body-lightening was indicated by the range, − 1 to − 4, with − 4 denoting the greatest extent of body-lightening. Final results for a number of experiments were averaged and are presented in tabular form in Table I. These results have been further analyzed so as to present the distribution of CDH and CBLH within the central nervous system

of each of the species considered (see Tables II and III). In these tables, the relative distribution of activity of the hormones is calculated for the various portions of the nervous system for each species.

This was done as follows. The average values of the chromatophores at 5, 10, 15, and 30 minutes following extract-injection were of themselves averaged. Then for Table II the portion of the nervous system producing maximum darkening was

TABLE II

The quantitative distribution of CDH activity within the central nervous systems of a number of crustaceans. The region of maximum activity is arbitrarily given the value 1.00. It is important to note that each portion of the nervous system, regardless of size, is extracted in an equal volume of sea-water, and the relative concentrations of the principles investigated are expressed solely in terms of their activities. This note applies equally to Table III.

Classification	Species or genus	Brain	Connectives	Thoracic cord	Abdominal cord
Isopoda	*Idothea baltica*	1.00	1.00	1.00	1.00
Decapoda					
Natantia	*Crago septemspinosus*	some 0.06	1.00	0.22	0.21
	Palaemonetes vulgaris	0.85	1.00	0.97	0.98
Reptantia					
Astacura	*Homarus americanus*	1.00	0.61	0.61	0.53
	Cambarus virilis	1.00	0.84	0.94	0.84
Anomura	*Upogebia affinis*	1.00	0.10	0.80	0.65
	Pagurus sp.	0.17	0	1.00	—
	Emerita talpoidea	0	0	1.00	—
Brachyura					
Oxyrhyncha	*Libinia* sp.	0	0	0	—
Brachyrhyncha	*Cancer irroratus*	0	0	0	—
	Carcinides maenas	0	0	0	—
	Ovalipes ocellatus	0	0	0	—
	Uca pugilator	0	0	0	—

arbitrarily given the value 1.00, the activity of the other parts being expressed in terms of simple proportions of this. For Table III the part showing maximum lightening was given the value − 1.00 with the activity of other parts similarly expressed proportionately. The positive values in the latter table obviously indicate darkening rather than lightening.

Within the single species of *Isopoda* investigated, *Idothea baltica*, there appears to be roughly a uniform distribution of CDH throughout the central nervous system, all organs darkening the telson and uropods of *Crago* to approximately the

same degree. Great variations in distribution of the hormones occur among the decapods. The Natantian, *Crago* apparently possesses significant CDH activity only in the regions of the circumoesophageal connectives. CDH is differentially distributed throughout the central nervous system of the anomurans with highest quantity usually in the posterior region of the thoracic cord, is relatively uniformly distributed within the central nervous system of the astacurans and *Palaemonetes,* and is entirely absent within that of the brachyurans.

The quantitative distribution of CBLH was considered here solely within the reptantian nervous system, although it is known to be present throughout the central nervous system of the natantians (Brown and Wulff, 1941). Both the anomurans and brachyurans show wide distribution of this principle throughout brain,

TABLE III

The quantitative distribution of CBLH activity within the central nervous systems of a number of crustaceans. The region of maximum body-lightening is arbitrarily assigned the value − 1.00. The + values indicate body-darkening.

Classification	Species or genus	Brain	Connectives	Thoracic cord	Abdominal cord
Isopoda	*Idothea baltica*	pres.	pres.	pres.	pres.
Decapoda					
Natantia	*Crago septemspinosus*	pres.	pres.	pres.	pres.
	Palaemonetes vulgaris	pres.	pres.	pres.	pres.
Reptantia					
Astacura	*Homarus americanus*	+2.40	−1.00	+2.20	+7.20
	Cambarus virilis	+1.09	−0.73	−1.00	+1.00
Anomura	*Upogebia affinis*	−0.64	−1.00	+0.27	−0.18
	Pagurus sp.	−0.92	−0.85	−1.00	
	Emerita talpoidea	−1.00	−0.86	−0.57	
Brachyura					
Oxyrhyncha	*Libinia* sp.	−1.00	−0.67	−0.71	
Brachyrhyncha	*Cancer irroratus*	pres.	pres.	pres.	
	Carcinides maenas	pres.	pres.	pres.	
	Ovalipes ocellatus	pres.	pres.	pres.	
	Uca pugilator	pres.	pres.	pres.	

connectives, and thoracic cord. However, a striking feature is noted in the astacurans and the natantian, *Palaemonetes,* in which a darkening (see Fig. 1*A*), as well as a lightening, of the body occurs.

The two species of astacurans with which we have concerned ourselves more or less parallel one another with respect to the distribution of CDH. In *Homarus* and *Cambarus* the region of greatest quantity of this principle is the brain, and is followed by an apparent gradual diminution of the substance from anterior to posterior within the nervous system. The problem of CBLH distribution seems somewhat more complex since, as has been previously mentioned, certain of these nervous-system extracts appear to produce body-darkening preceded by a body-lightening. The abdominal-cord extract is particularly active in body-darkening and only the

connectives and thoracic cords of *Homarus* and *Cambarus* show any body-lightening activity at all. In these cases where body-lightening is indicated, the lightening persists for only a short time and is followed by a definite darkening. These observations suggest that the body-darkening activity observable for extracts of the astacuran central nervous system is explainable in terms of CDH. It is significant that in no case is body-darkening ever obtained from a portion of the nervous system lacking tail-darkening activity. However, since there is no essential direct correlation between the degree of tail-darkening and the degree of body-darkening even within a single species, the observed results must be the consequences of varying proportions of the two principles within the extracts, with the degree of influence of either one being a function of its relative concentration at any given instant.

There are significant differences in the distribution of CDH within the group of anomurans. *Pagurus* and *Emerita* exhibit similar tail-darkening activities and these are shown chiefly by thoracic cord extracts. On the other hand, extracts of

Table IV

The responses of eyestalkless Crago to injections of extracts of parts of the thoracic cord of some anomurans, showing the differing distributions of CBLH and CDH activity. No. of cases signifies number of donors.

			Tail-darkening Time (min.)							Body-lightening ⊖ or darkening ⊕ Time (min.)						
Species	Part of thor. cord	No. cases	0	5	10	15	30	45	60	0	5	10	15	30	45	60
Pagurus pollicaris	Anterior ¼	8	0.0	0.3	0.3	0.4	0.3	0.0	0.0	0.0	−2.3	−2.2	−2.0	−0.6	−0.3	0.0
	Second ¼	8	0.0	0.6	0.7	0.7	0.2	0.0	0.0	0.0	−0.7	−0.6	−0.3	−0.3	0.0	0.0
	Third ¼	8	0.0	1.5	1.6	1.6	0.5	0.5	0.4	0.0	−0.4	−0.4	−0.3	0.0	0.0	0.0
	Posterior ¼	8	0.0	2.6	2.6	2.4	0.9	0.5	0.0	0.0	−0.5	−0.5	−0.3	−0.1	0.0	0.0
Pagurus longicarpus	Anterior ¼	6	0.0	0.5	0.5	0.5	0.0	0.0	0.0	0.0	−1.7	−1.6	−1.2	−0.4	−0.2	0.0
	Second ¼	6	0.0	0.2	0.2	0.2	0.0	0.0	0.0	0.0	−1.0	−1.0	−0.3	0.0	0.0	0.0
	Third ¼	6	0.0	1.3	1.4	1.5	0.8	0.5	0.2	0.0	−1.2	−0.8	−0.2	0.0	+0.2	+0.2
	Posterior ¼	6	0.0	2.0	2.0	2.0	1.2	0.5	0.0	0.0	−0.5	−0.4	−0.2	−0.2	0.0	0.0
Emerita talpoidea	Anterior ⅕	4	0.0	0.8	0.8	0.8	0.3	0.0	0.0	0.0	−2.3	−2.3	−1.8	−0.5	0.0	0.0
	Second ⅕	4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	Third ⅕	4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	−0.3	−0.3	0.0	0.0	0.0	0.0
	Fourth ⅕	4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	−0.5	−0.5	0.0	0.0	0.0	0.0
	Posterior ⅕	4	0.0	0.3	1.8	1.8	0.3	0.1	0.0	0.0	−0.3	+0.8	+0.8	0.0	0.0	0.0

brain, thoracic and abdominal cords of *Upogebia* all contain notable amounts of CDH. Another similarity between extracts from *Pagurus* and *Emerita* is seen in the distribution of CBLH. CBLH is found in considerable amounts in brain, connectives, and thoracic cord of both genera. However, the thoracic cord extracts of *Upogebia* show almost a complete absence of CBLH activity while the extracts of the remaining parts of the central nervous system produce a definite body-lightening, the connectives being most active in this respect.

The absence of CDH within the brachyurans investigated as well as the restriction of this principle to the connective region of the natantians studied confirms the results of Brown and Ederstrom (1940). Experimental data show that moderate amounts of CBLH are found in brain, thoracic cord, and connectives. Although results for CBLH distribution for brachyurans are shown only for *Libinia,* it has been found that they are qualitatively the same for *Uca, Cancer, Carcinides,* and

Ovalipes. The striking body-lightening effect of a strong extract of *Uca* connectives and commissures is illustrated in Figure 1*B*.

An attempt was made to analyze further the localization of CDH and CBLH within the thoracic cords of *Emerita* and two species of *Pagurus: pollicaris* and *longicarpus* (Table IV). The procedure consisted of dividing the thoracic cords into a number of approximately equal portions, four in the case of *Pagurus* and five in that of *Emerita*. It was observed that the concentration of CDH within the thoracic cord of both *P. pollicaris* and *P. longicarpus* is greatest in the posterior fourth of the cord and decreases gradually along the cord as one proceeds anteriorly. In *Emerita* the highest region of CDH concentration is also the posterior portion of the thoracic cord. However, there is a lack of CDH in any of the central portions of the thoracic cord in *Emerita*. It would seem then that the distribution of CDH in the thoracic cord of *Emerita* is more restricted than in *Pagurus*.

The distribution of CBLH in the thoracic cord of *P. pollicaris* and *P. longicarpus* is similar. The most intense body-lightening effect is brought about by extracts of the anterior fourth of the cord while less intense reactions are produced by extracts of the remaining portions. Experiments with extracts of *Emerita* thoracic cord indicate a higher concentration of CBLH in the anterior portion of the cord, and apparent absence of CBLH in the second portion and only slight amounts of the principle in the third, fourth and fifth divisions of the cord. In summarizing the distribution of CDH and CBLH within the thoracic cords of *Pagurus* and *Emerita* we can say that CDH is relatively more concentrated posteriorly in the thoracic cord while CBLH appears more concentrated anteriorly.

Discussion of Results

The effect of the extracts of the central nervous system upon the dark pigments of the telson and uropods of *Crago* possesses a characteristic pattern in each of the major groups of the order Decapoda. In the Natantian, *Crago*, we have observed the restriction of CDH activity to the circumoesophageal connectives, whereas the *Astacura* and *Palaemonetes* exhibit a more generalized occurrence of the hormone within the organs of the central nervous system. However, as one proceeds to the *Anomura*, these contain changes from the widespread condition in the astacurans to a more specialized one as evidenced by the restriction of CDH in the thoracic cord of two of the three genera examined. Finally there is an entire lack of CDH among the brachyurans.

Experimental data concerning the distribution of CBLH in the reptantians present an interesting problem. Although both the anomurans and brachyurans possess the body-lightening hormone in varying amounts throughout the entire central nervous system, the astacurans appear to limit the hormone to connectives and thoracic cord. The simplest explanation for the body-darkening activity of the astacuran central-nervous-system extracts involves action of the tail-darkening principle. It is thought that CDH produces body-darkening after CBLH has been exhausted or in the absence of CBLH. This is indicated in Figure 2 in which selected portions of the central nervous system of *Libinia*, *Cambarus*, and *Homarus* are shown to produce a graded series of differential effects upon the coloration of the body of eyestalkless *Crago*. These range all the way from maximum body-lightening and no trace of darkening (*Libinia* brain) through initial lightening followed by

darkening, to immediate and extensive body-darkening (*Homarus* abdominal cord). These results are believed to be explained in terms of different relative amounts of CDH and CBLH. The former is known to be absent in the case of *Libinia*, and it is assumed that the latter is absent or nearly so in the case of *Homarus* abdominal cord. In the case of the extracts of *Homarus* thoracic cord and connectives and those of *Cambarus* thoracic cord, CBLH is present in small amounts and lightens the body for a short time, thereby delaying the darkening influence of CDH on the body.

A comparison of tail-darkening and body-darkening within *Crago* injected with nervous system extracts from numerous sources suggests a rough positive correlation between the two (Fig. 3). Generally speaking, we may infer from these data that the tendency towards body-darkening is greater in those animals showing a high degree of tail-darkening. This gives further support for an active role of CDH in body-darkening.

Unlike the *Decapoda* the *Isopoda* apparently exhibit a uniform distribution of CDH within the central nervous system. However, since only a single species was considered, further experimentation is deemed necessary before any decisive statement is made concerning CDH distribution within this group.

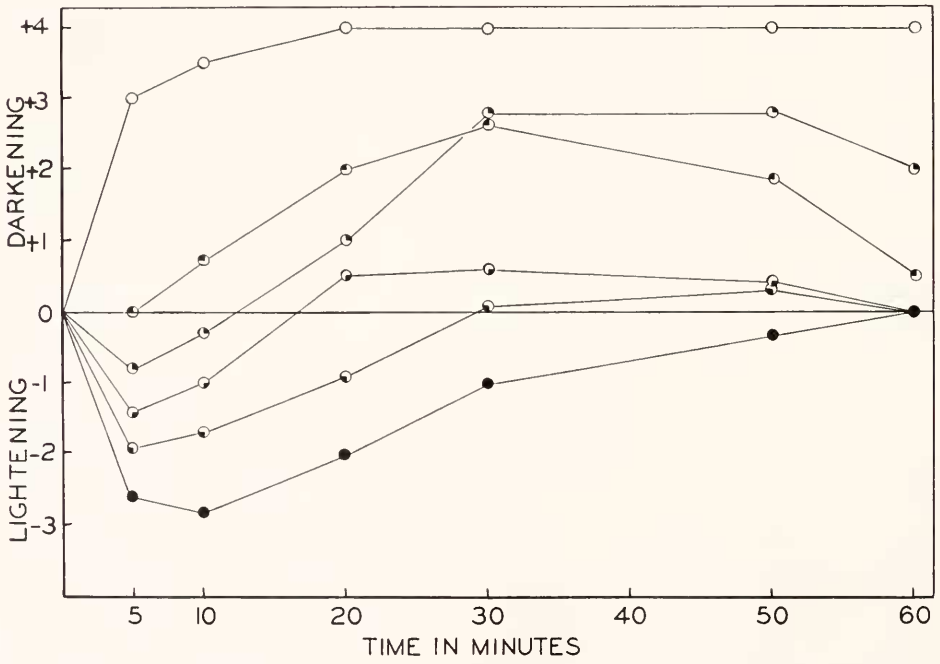

FIGURE 2. The influences of extracts of selected portions of the central nervous system of some crustaceans upon the body coloration of eyestalkless *Crago*.

From most positive to most negative at the end of 10 minutes are shown, respectively, *Homarus* abdominal cord, *Homarus* brain, *Homarus* thoracic cord, *Homarus* circumoesophageal connectives, *Cambarus* thoracic cord, and *Libinia* brain. Concentration in each experiment was: organs of one specimen/0.5 ml. sea-water.

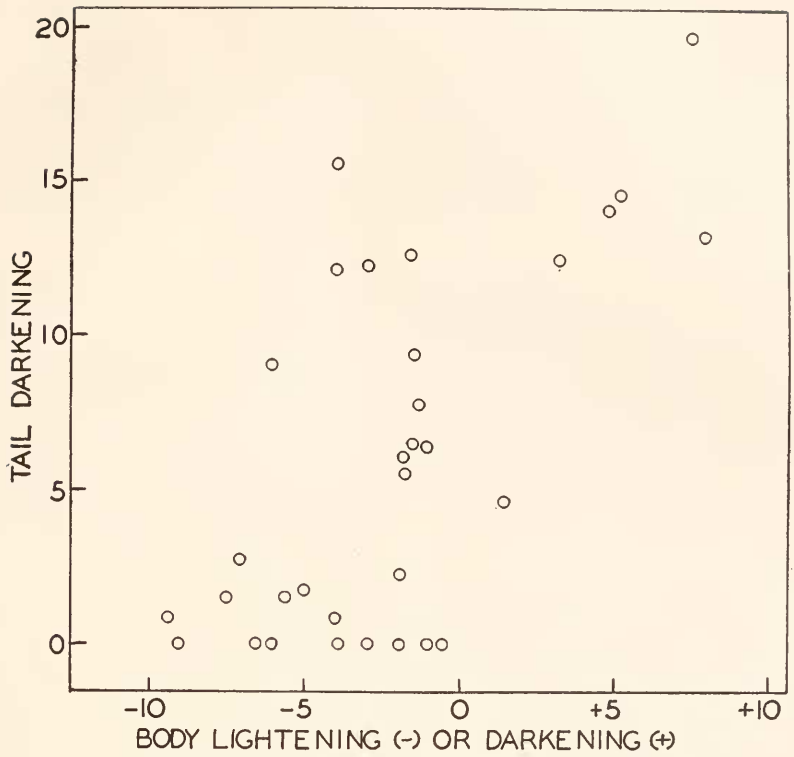

FIGURE 3. The general relationship between the degree of darkening or lightening of the body proper of eyestalkless *Crago* and the degree of darkening of the telson and uropods. Darkening of the tail is expressed as the algebraic sum of the intensities of the reactions at 5, 10, 15, 30, 45, and 60 min. following extract injection, thereby including a measure of both intensity and duration of the effect. Body-lightening, being more rapidly transitory, is expressed as the algebraic sum of the values at 5, 10, 15, and 30 min.

SUMMARY

1. A survey was made of the effects upon *Crago* color-change of sea-water extracts of various parts of the central nervous system of thirteen species of higher crustaceans. The crustaceans represented the groups *Isopoda, Natantia, Astacura, Anomura,* and *Brachyura.*

2. Extracts of various portions of the nervous system among the various groups showed wide differences in their total chromatophorotropic activities, producing various degrees of telson and uropod darkening and of body-lightening and darkening.

3. An analysis of the results gave support to the hypothesis that most crustacean nervous systems possess at least two principles, *a*) a *Crago* body-lightening principle, CBLH, lightening all portions of the body except telson and uropods, and *b*) a *Crago*-darkening hormone, CDH, darkening the telson and uropods, and, in the absence of CBLH, the body as well.

4. CBLH is more or less uniformly distributed throughout the nervous systems of all the species examined except the astacurans in which it is demonstrated only for the circumoesophageal connectives and thoracic cord.

5. CDH is restricted to the circumoesophageal connective region of the *Natantia,* is differentially distributed throughout the nervous systems of anomurans, with highest concentration in the posterior region of the thoracic cord, and is distributed throughout the nervous systems of the other species except the brachyurans in which it is absent.

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